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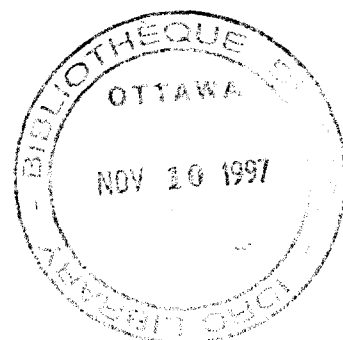
Tagging and marking fish for IDRC research projects

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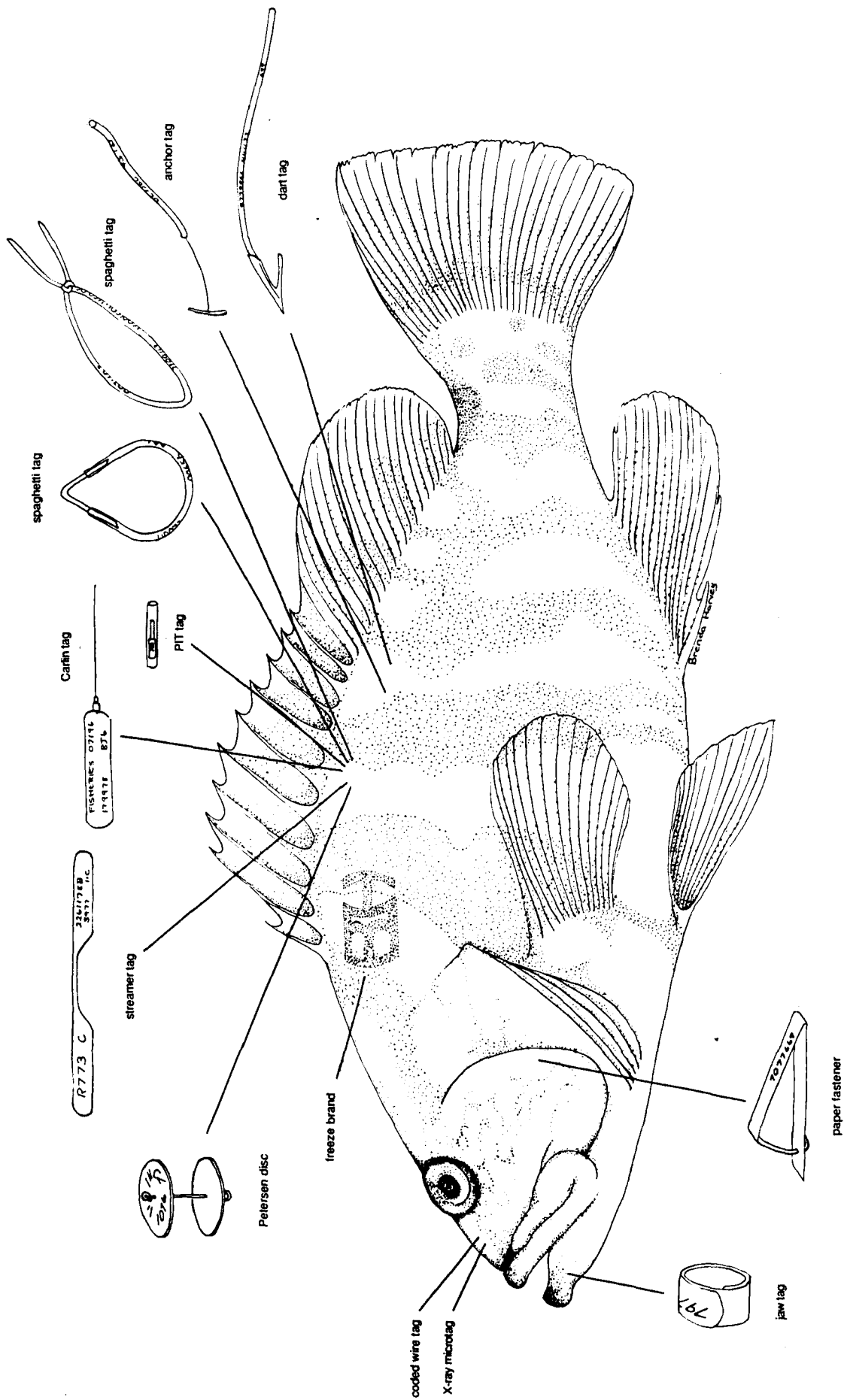
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Tagging and marking fish for IDRC research projects

Introduction

Anybody attempting to learn about fish marking and tagging techniques will be confronted by a very large published literature and one or two recent reviews. From the standpoint of most IDRC Fisheries projects, much of the published information is irrelevant, because the overwhelming drive behind development of tagging and marking methods has been to enable marking of large numbers of fish in batch lots for mark-recapture, stock assessment and migration studies. These population-oriented studies are usually carried out by Governmental Fish and Wildlife agencies, commonly involve the participation of commercial and sport fishermen in collection of tagged fish, and have generated an entire subset of mathematically oriented studies dealing with the measurement of tag shedding rates.

The tags and marks developed for large-scale population studies reflect the following needs: the need to mark large numbers of fish cheaply and rapidly and without a requirement for great skill; the need to include contact instructions on the tag; the need for a tag or mark that does not interfere with normal behaviour in the wild and does not make the animal more susceptible to catching or predation; and the need for high visibility in cases where tags must be spotted from a distance.

For most IDRC research projects these needs are relatively unimportant, and much more can be gotten out of the tagging literature by trying to determine why certain kinds of tags fail, and from there defining a set of principles of tag design that can be adapted to a particular experimental situation. Fish have been tagged since the late nineteenth century, and many reviews still include discussions of tags that have hardly changed in fifty years. Yet there have been major advances in tagging technology, particularly in the development of electronic methods, and it is the aim of this

brief review to allow the fisheries researcher working on an IDRC project to gain an overview of the advantages and disadvantages of all of these tag types that is uncomplicated by criteria that have no meaning for his study.

Requirements of a fish mark or tag in an IDRC study

Most IDRC studies are carried out in controlled situations where fish are held in captivity in tanks, net cages or ponds. The numbers of fish tagged are more likely to be in the hundreds than in the thousands; the requirements for batch marking (a group of individuals receiving the same identifying mark) and individual marking (each individual receives a unique number or code) are probably evenly split. Because plenty of skilled manpower is usually available there is no great requirement for speed or particular ease of application. Tag retention should be in the six month to one year range; a tag that would be rejected for a long term population study in the wild may be perfectly acceptable for a smaller scale study in which fish are handled periodically and can be remarked. Cost and availability are important considerations in Third World studies, yet cost in particular should be carefully looked at to avoid false economies. Some of the newer electronic tags are expensive on an individual basis, but the value of the fish and whatever treatments it may have received must be considered. There is no logic in applying \$ 5-10 in hormone treatment to a fish tagged with a 20 - cent tag that falls out after a month.

There are presently tags available to suit the requirements of most studies. To select the right one, the researcher should consider the following:

Size of fish

Small fish are likely to grow, so that external marks or brands are expanded or obscured, and external tags engulfed. Tags that rely on anchoring behind bony fin ray extensions must be

carefully chosen with regard to the spacing of these rays (see below for a discussion of the two most important external tags, dart and anchor tags, in relation to fish size).

Integument

Scale type and size varies enormously among fish. A fish with an abrasive skin - a shark is an extreme example- will cause a plastic external tag to wear through. Cold branding is also influenced by scale size, with generally better results obtained by small scaled fish.

Handling stress

Many tags fail because of ulceration and disease at the point of application. The type of mark or tag can only partially be blamed; it is equally important to stress fish as little as possible during the handling required for tagging. A fish tagged with a minimum of capture-related stress is less likely to develop a debilitating wound at the tagging site.

Tag quality

External and internal tags are made by several manufacturers (see Appendix 1), who also supply associated equipment such as tagging guns. Tags can suffer from separation of external and internal portions, illegibility and insufficient accompanying records, and users should consult their colleagues where possible to avoid buying a tag with a well-known problem.

Types of marks and tags available

Fish can be marked (by dye injection, branding, tattooing, spray painting, fin clipping, oxytetracycline and microtaggant injection), tagged with external or internal mechanical tags (dart and anchor tags, streamers, clips and discs), implanted with sophisticated internal tags (PIT tags, X-ray microtags, coded wire tags) or identified by characteristic "fingerprints" (blood groups, meristic characteristics, scale circuli).

Marks

The best external marks are made by fin clipping, branding and jet injection. All these methods suffer from the disadvantage common to all marking methods, namely the limited amount of information that can be contained in each mark; however, individuals can be identified within limits set by the number of available locations for marks.

Fin clipping and opercular punching

Complete removal of the adipose fin is a well tested method of identification but serves only for batch marking or to alert to the presence of an internal tag. Clipping of all other fins, unless done at point of attachment to bone, results in some degree of regeneration, depending on the fish species and how close to the body the cut was made. Nevertheless, irregularities at the site of regeneration have some use as identifying marks. Partial clips are useful for studies of a few weeks duration at best, and holes or nicks made in fins fill in within days. Fish such as siganids with protruding dorsal spines can be marked individually using a binary system, although strong clippers are needed to cut through the bone. When pectoral fins are excluded for reasons of swimming efficiency, there are only ten possible combinations of two-fin marks.

Holes punched in opercula (using a paper or leather punch or modified pliers) are suitable for short-term studies only, and are most successful in fishes with bony opercula. With larger fish, however, the size of the operculum means that a reasonable number of codes dependent on number and location of holes can be worked out.

Branding

Some success has been reported with hot brands and brands drawn with silver nitrate pencils, but the most important technique uses a branding tool cooled in liquid nitrogen and applied to the dorsal integument for approximately two seconds. The great advantage of cold branding is that it is noninvasive and does not affect fish behaviour; the main disadvantages are fading and the lack of information that can be carried on a brand. Number codes require a set of numbered brands, and will become illegible as fish grow; in actual practice the symbols used are simple shapes that are repeated, placed in different locations or combined with other marks such as fin clips to generate more information.

Success of branding depends greatly on scale size; fish with large, thick scales should be branded in areas where scales are reduced or absent. The small scales of juvenile fish generally make them much easier to brand than adults. The important thing to remember is that brands remain legible for two reasons: there is a primary effect in which dark-colored melanin-containing cells invade the branded area, and a secondary effect in which regenerating scales in the branded area are smaller and deformed. This secondary effect can often be seen long after the primary darkening effect has faded, particularly if the fish is viewed at an angle.

Branding tools are usually hand made. Copper is widely used, as it is easily bent and has a high thermal capacity; silver and stainless steel have also been used, with better results with silver; neither would appear to have any advantage over copper.

Freeze brands can be recognizable for more than a year, depending on the species and the age at branding. An excellent way to maximize legibility is to rebrand periodically, say every six months; in many studies fish are handled at least this often, and the branding process is simple and rapid. With rebranding and limited numbers of fish to be identified, and assuming a supply of liquid

nitrogen to be available, freeze branding is a simple, inexpensive and effective marking technique.

Jet Injection

Stains and dyes applied with sufficient force to the outer surface of a fish will lodge in the epidermis and dermis and remain highly visible for long periods. A number of marking techniques are based on this effect, including injection of dyes, spray painting and tattooing (see below). The best application for individual marking of relatively small numbers of fish is by jet injection, in which a hand-held dental tool ("jet inoculator") is applied to the surface of the fish and a small amount of Alcian Blue stain in solution forced into the skin as a discrete dot. Marks should be made directly on fin rays or on the lighter coloured underbelly, where the area available leaves scope for devising individual codes. As with freeze branding and all other marking techniques the duration of mark retention varies, but periods of 6 months to a year are common, and periodic remarking is easy. If dots are kept small a number of combinations of fin and body locations can be used to identify individuals, although, as with fin clipping and branding schemes, complicated codes rapidly become self-defeating in field situations.

Tattoos, dye Injection, spray marking

These techniques all depend on deposition of pigment particles in or under the dermis; the exact placement varies with the technique and with the skill of the operator. Tattooing is not popular now, requiring as it does a tattooing device and offering no real advantages over jet injection. Hand injection of pigments in or under the dermis in lightly pigmented areas of fish has been widely used with latex emulsions and acrylic polymer emulsions sold as artists colours, with results visible for over a year. Guidelines for application are of little use because of the differing response of different species and the dependence of retention on paint manufacturer and colour shade;

anyone wishing to try this method should simply purchase a few tubes of artists' acrylic colours and experiment in the belly and jaw area using the smallest gauge needle that will readily expel the emulsion.

Spray marking is another technique based on uptake of dye particles, this time applied at high pressure using a paint sprayer or similar device held a foot or so away from the fish. The technique is useful only for applying a batch mark, and has its greatest application in rapidly marking large numbers of fish. It has been used mostly with fluorescent pigments applied to young salmonids, where mortality is surprisingly low and retention in the order of several years. A disadvantage of the technique is the requirement for ultraviolet illumination to see the pigment deposits in marked fish.

Microtaggants

Microtaggants are microscopic color-coded plastic particles originally developed to identify batch lots of explosives and to mark tools and equipment. Particles suspended in physiological saline have been applied to the integument of fish using a jet inoculator; after location under ultraviolet light, the color-coded laminations are read under low magnification.

Oxytetracycline marking

The antibiotic oxytetracycline is fixed in the bone of growing animals and will fluoresce under ultraviolet light. Addition of low levels of the drug to feed will result in marked vertebrae, and some estimate of growth rate can be inferred from the position of the mark. The method has been used mostly for salmonids, with some field application for marking sharks and large pelagic fish via intramuscular injection; the chief drawback for use in aquaculture is the potential for developing resistance to the antibiotic.

External or Internal mechanical tags

The great advantage of mechanical tags is their ability to bear coded information so that large numbers of individual fish can be recognized. Most of the commercially available and popular tags fall into this category, and prices per tag are low. Once the principles of tag retention are understood, particularly the dependence on size of fish, there is much scope for individual design, and many handmade tags have been used successfully. The main disadvantage of mechanical tags is the need to attach them through muscle or bone, and the resulting infections that can be produced by abrasion and drag.

Anchor and dart tags

Anchor and dart tags are currently the most widely used external tags. Both depend on proper implantation and will quickly be lost if applied incorrectly.

Both are available in several sizes and from several manufacturers.

A dart tag consists of a V-shaped plastic head molded or glued to a tubular plastic tail that bears a unique inscription or code. Anchor tags differ in that the head region is T-shaped, and resemble the tags used to attach prices to clothing. Dart tags are inserted using a hand-held hollow needle; for anchor tags the insertion process is analogous but automated by the use of a tagging gun that dispenses the tags from a magazine.

Both types of tag function properly only if the barb or T-bar locks securely behind the bones (pterygiophores) supporting the dorsal fins; neither will hold well in muscle, although one tag manufacturer is developing a modified dart tag intended to function this way. This requirement for penetration and locking between pterygiophores dictates the size of tag to be used: the

length of tag and the length of the needle on the tagging gun must be selected with the size of the fish in mind. As a general rule, anchor tags are more successfully used with smaller fish, because the slender T-bar can pass easily between pterygiophores, and because they can be applied with a smaller needle. However, the only reliable way to choose the proper tag type and size is to partially dissect a specimen, implant a number of trial tags, and verify that the tag configuration and size work well with the anatomy of the particular fish. Implanting a too-large dart tag in a small fish will break the pterygiophores, while a too-small anchor tag will simply slip back out between them.

notes?
Anchor and dart tags cause wounds that can become inflamed and lead to mortality in the tagged population. The best way to avoid such complications, as with all invasive tagging methods, is to ensure that handling stress is minimised through the use of anesthetics and adequate recovery facilities.

Discs and clips

There is a wide variety of tags that are wired or pinched onto various parts of fish. The oldest is the Petersen disc, which consists simply of two plastic discs held to the sides of the fish by a metal pin passing through the flesh beneath the dorsal fin. Problems with chronic lesions caused by friction can often be minimised by relieving the underside of the disc or by custom-fabricating discs with holes for two attachment pins. The principle of the Petersen disc can easily be applied to any number of homemade tags.

Dangler tags consist of a small plastic plate wired to the fish; the most commonly used is the Carlin tag. This tag must be applied using a pair of hypodermic needles, with requirements for time and dexterity. Dangling tags can easily be snagged in nets.

Jaw tags are metal rings resembling bird bands and have been used successfully so long as they are not large enough to interfere with feeding. They can be obtained, sequentially numbered, from manufacturers of general fastening equipment. Enterprising researchers will even find that small paper fasteners can be crimped onto opercula and retained for several months; the fasteners should be painted to prevent corrosion and can be numbered serially.

The term "spaghetti tag" is commonly misused to refer to any tag having a vinyl tubular tail. In fact a spaghetti tag is a length of vinyl tubing bearing a legend or code and threaded through the dorsal musculature of a fish. A long spaghetti tag is knotted and the ends left to trail in the water; the ends of shorter spaghetti tags can be held together with special inserts so that the tag forms a circle. Lock-on or cinch-up tags are variants available from major manufacturers. Spaghetti tags are applied using a hollow needle and are highly visible; however, there is evidence that fish tagged this way are more susceptible to predation, and drag frequently causes wounds that can become infected. For large fish in a short term study, the method is effective.

Another type of tag that is threaded through the dorsal musculature is the streamer tag. These can be purchased from fish tag manufacturers in various sizes and complete with needles for insertion. However, they can also be made cheaply from vinyl surveyors' or "flagging" tape upon which numbers are written with indelible pen. The tag is threaded onto a needle and pulled through the fish until the narrow part is in the dorsal musculature. The method has not been widely reported, but its simplicity holds promise for adaptation to a number of species.

Internal tags: coded wire, X-ray and PIT tags

Intraperitoneal or intramuscular implantation of small plastic and metal tags has been practiced with some success since the 1930s; obvious advantages are lack of interference with behaviour and

the ability to record individual information; a major disadvantage is the need for a detector or other means of alerting researchers to the presence of a tag.

The application of electronics to tagging technology has capitalized on the advantages of implantable tags while eliminating many of the disadvantages. The two most important systems are coded wire tags (and the variant X-ray microtag) and passive inductive transponder (PIT) tags. Both are considerably more expensive than mechanical tags.

Coded wire and X-ray microtags

Coded wire tags are small (less than 1 mm) lengths of wire carrying information in the form of notches cut into the tag and implanted using specialized automated equipment. The tags cannot now be used for individual recognition; a typical use is for identifying large numbers of salmonids released from hatcheries. Tilapias have also been tagged using the system, but for any other than a very large scale study the method is uneconomical (marking equipment cost approximately \$25,000 USD in 1987). Tags are detected by a metal detector or by an external mark (removal of the adipose fin is common). A newer version of the coded wire tag is the internally readable X-ray microtag; this tag need not be dissected from the fish for reading, but can instead be read without harm to the fish by X-raying the animal. The same considerations of cost and non-individuality of numbering apply. The manufacturer of coded wire tags is presently developing systems that will allow individual identification of fish, and that require less costly apparatus for implantation.

PIT (Passive Inductive Transponder) tags

A PIT tag is about the size of a grain of rice and contains a microprocessor chip and antenna. Each tag is individually coded. When the tag is excited by a hand-held or tunnel detector, it transmits its unique code to a reading device which automatically decodes, displays and stores it. PIT tags are

implanted intramuscularly or intraperitoneally using a hollow needle, and have been extensively used for a number of fish and wildlife applications. For small scale fisheries experiments, detection and display of the code is done using a hand-held reader.

The advantages of PIT tags are small size and consequent high retention in small and large fish, lack of interference with normal behaviour, and virtually limitless available individual codes. The most obvious disadvantage is cost: hand-held readers cost approximately \$2225 USD in 1987, with the tags themselves costing \$5.60 USD each. Nevertheless, such an outlay is probably justified in situations where several groups can use the equipment and considerable time, effort and expense have gone into the husbandry and treatment of experimental fish.

Biological "fingerprints"

There are some existing morphological or biochemical characteristics of fish that can be used to separate stocks; these can be useful in studies where population sizes are large. Such characteristics have a genetic or an environmental component or both, and their interpretation must be mathematically based. Meristic characters such as the number of gill rakers, fin rays or scale circuli have been used to separate different stocks of the same species, as have pigmentation and body size and shape.

Because morphological characteristics are influenced by the environment, a better way of separating stocks, races or species of fish is to look at more direct expressions of genetic differences on a biochemical level. The presence or absence of a number of proteins in a variety of tissues including blood and muscle can be detected through electrophoresis, and the technique allows distinction of subtle differences between populations by looking at genetic variation over a number of loci. In studies where identification even at the species level is difficult

ack of individual mark
and where knowledge of parentage is vital for breeding and genetic conservation purposes, electrophoretic analysis of tissue proteins is indispensable.

Future applications of electrophoretic techniques will probably include true genetic tagging, in which unique alleles with readily recognizable but harmless external expression can be deliberately bred into study populations.

Recommendations

Fisheries studies vary enormously in numbers of animals, objectives, species and facilities available, so recommendations are difficult. Some general guidelines:

Fin clips and cold brands. These methods are cheap (assuming liquid nitrogen is available) and very easy. They are good for small scale studies where periodic remarking is feasible. A drawback is the tendency to devise over-complicated code schemes that end up by confusing everyone. Individual identification is possible for a limited number of fish only.

Anchor and dart tags. These methods are probably the most popular today and will give excellent results for all sizes of study so long as tag type and size are carefully chosen. Individual identification is possible for an unlimited number of fish.

PIT tags. These tags should be considered for highly reliable individual identification without disturbance to behaviour. Initial cost is high but probably supportable for large continuing studies and in cases where detection equipment can be used by several groups.

Check anatomy first. If contemplating an external mechanical tag, dissect a typical specimen and look for solid muscle attachment sites and pterygiophore spacing. This is essential before

ordering dart or anchor tags. Try a tag borrowed from someone else or make a mock-up so that you don't end up with a size or shape that doesn't work.

Experiment. There are many successful tagging methods that were tailor-made for the particular fish and study; the diversity of fish sizes and shapes often demands this. Secure attachment and minimal abrasion are the most important characteristics of a mechanical tag, and something can often be rigged up with local materials.

Duration of study and remarking. For studies lasting only a few months, simple marking techniques can always be repeated every time the fish is handled. Freeze branding and jet injection of pigments are excellent examples.

Buy quality. When purchasing manufactured fish tags, consult with colleagues whenever possible to find out whether they have had any problems with a particular brand. Tags are made by a variety of glueing and molding methods; glueing in particular is an unreliable means of attaching parts together and has been responsible for many tag failures.

Stress and disease. Most tagging and marking methods apart from freeze branding are invasive. Cuts or punctures should be treated prophylactically with an antibiotic cream. More important, fish should be anesthetized for all handling procedures. 2-phenoxyethanol is a rapidly acting and relatively forgiving sedative when applied as a bath (50-100 mg/l). A newer anesthetic, metomidate hydrochloride (Marinil) is also highly effective as a bath (1-4 mg/l); it can also be used as a strong solution (20 mg/ml) squirted into the mouth of a fish caught in a net but not yet removed from the water. A few millilitres applied this way and flushed over the gills will produce very rapid knockdown with minimal water contamination, and fish need not be removed from the water until sedated.

Appendix 1. Sources of tagging and marking materials

Hallprint Pty. Ltd.,

27 Jacobsen Crescent,

Holden Hill, S. Australia 5088

High quality dart, anchor, loop, internal, wire-on, streamer and other mechanical tags for fish, crustaceans and molluscs. Custom tags can be developed along with user.

Floy Tag and Manufacturing Inc.,

4616 Union Bay Place Northeast,

Seattle, Washington 98105.

Manufacturers of mechanical fish tags.

Sigma Chemical Company,

P.O. Box 14508,

St. Louis, Missouri 63178

Biological stains and chemicals

National Band and Tag Co.,

721 York St.,

Newport, Kentucky 41072

General fastenings, paper fasteners etc.

**Safety and Security Systems Division,
223-3N 3M Center,
St. Paul, Minnesota 55144.**

Microtaggants

**Northwest Marine Technology Ltd.,
Shaw Island, Washington 98286**

Coded wire tags.

**Biosonics Inc.,
4520 Union Bay Place NE,
Seattle, Washington 98105**

PIT tags.

**Wright Health Group Ltd.,
Kingsway West, Dundee DD2 3QD,
Scotland**

Panjet dental inoculator (needleless injector). Manufacturer supplies extensive literature on applications.

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